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<120> Biosensor materials and methods

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<141> 2000-03-14

<150> PCT/GB98/01893
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agtgtcggac agagcgccga acacgaggta gaggaggacg gcgacgatgc cggcgacgaa 5340
tactccggcc agggcgaagg tgcggctgat ccccgcgacg gtggtgccgt aggagacgag 5400
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```

```
cttctcgcct gccgcatagc gcccttcgag cagccggttc ttcagaatgt cgtacaacgc 7200
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cggcttgggg ccggtgtcgg tggtggtcat cgagtcaggc tttccgtcgg cgtcccggac 7320
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```

<210> 3
<211> 246
<212> PRT
<213> Rhodococcus corallina

<400> 3

```
Met Thr Thr Thr Asp Thr Gly Pro Lys Pro Gly Ser Glu Ala Ala Ala
  1               5                  10                  15

Leu Leu Ala Asn Val Arg Thr Ser Gly Ala Arg Leu Ser Ser Ala Leu
            20                  25                  30

Tyr Asp Ile Leu Lys Asn Arg Leu Leu Glu Gly Arg Tyr Ala Ala Gly
        35                  40                  45

Glu Lys Ile Val Val Glu Ser Ile Arg Gln Glu Phe Gly Val Ser Lys
    50                  55                  60

Gln Pro Val Met Asp Ala Leu Arg Arg Leu Ser Ser Asp Lys Leu Val
65                  70                  75                  80

His Ile Val Pro Gln Val Gly Cys Glu Val Val Ser Tyr Ala Pro Arg
                85                  90                  95

Glu Val Glu Asp Phe Tyr Thr Leu Phe Gly Gly Phe Glu Gly Thr Ile
            100                 105                 110

Ala Ala Val Ala Ala Ser Arg Arg Thr Glu Ala Gln Leu Leu Glu Leu
        115                 120                 125

Asp Leu Ile Ser Ala Arg Val Asp Ala Leu Ile Thr Ser His Asp Pro
    130                 135                 140

Val Val Arg Ala Arg Gly Tyr Arg Val His Asn Arg Glu Phe His Ala
145                 150                 155                 160

Ala Ile His Ala Met Ala His Ser Arg Ile Met Glu Glu Thr Ser Gln
                165                 170                 175

Arg Met Trp Asp Leu Ser Asp Phe Leu Ile Asn Thr Thr Gly Ile Thr
            180                 185                 190

Asn Pro Leu Ser Ser Ala Leu Pro Asp Arg Gln His Asp His His Glu
        195                 200                 205

Ile Thr Glu Ala Ile Arg Asn Arg Asp Ala Ala Ala Ala Arg Glu Ala
    210                 215                 220
```

```
Met Glu Arg His Ile Val Gly Thr Ile Ala Val Ile Arg Asp Glu Ser
225                 230                 235                 240

Asn Ala Gln Leu Pro Ser
                245
```

<210> 4
<211> 451
<212> PRT
<213> Rhodococcus corallina

<400> 4

```
Met Ala Ser Phe Ile Gly Thr Thr Val Glu Tyr Tyr Asp Phe Phe Ile
  1               5                  10                  15

Tyr Gly Thr Ala Ala Ala Leu Val Phe Pro Glu Leu Phe Phe Pro Asp
            20                  25                  30

Val Ser Ser Ala Ile Gly Ile Leu Leu Ser Phe Ala Thr Phe Ser Val
        35                  40                  45

Gly Phe Leu Ala Arg Pro Leu Gly Gly Ile Val Phe Gly His Phe Gly
    50                  55                  60

Asp Arg Val Gly Arg Lys Gln Met Leu Val Ile Ser Leu Val Gly Met
65                  70                  75                  80

Gly Ser Ala Thr Val Leu Met Gly Leu Leu Pro Gly Tyr Ala Gln Ile
                85                  90                  95

Gly Ile Ala Ala Pro Ile Leu Leu Thr Leu Leu Arg Leu Val Gln Gly
            100                 105                 110

Phe Ala Val Gly Gly Glu Trp Gly Gly Ala Thr Leu Met Ala Val Glu
        115                 120                 125

His Ala Pro Thr Ala Lys Lys Gly Phe Phe Gly Ser Phe Ser Gln Met
    130                 135                 140

Gly Ala Pro Ala Gly Thr Ser Val Ala Thr Leu Ala Phe Phe Ala Val
145                 150                 155                 160

Ser Gln Leu Pro Asp Glu Gln Phe Leu Ser Trp Gly Trp Arg Leu Pro
                165                 170                 175

Phe Leu Phe Ser Ala Val Leu Ile Val Ile Gly Leu Phe Ile Arg Leu
            180                 185                 190

Ser Leu Ala Glu Ser Pro Asp Phe Ala Glu Val Lys Ala Gln Ser Ala
        195                 200                 205

Val Val Arg Met Pro Ile Ala Glu Ala Phe Arg Lys His Trp Lys Glu
    210                 215                 220

Ile Leu Leu Ile Ala Gly Thr Tyr Leu Ser Gln Gly Val Phe Ala Tyr
225                 230                 235                 240
```

```
Ile Cys Met Ala Tyr Leu Val Ser Tyr Gly Thr Thr Val Ala Gly Ile
                245                 250                 255

Ser Arg Thr Phe Ala Leu Ala Gly Val Phe Val Ala Gly Ile Val Ala
            260                 265                 270

Val Leu Leu Tyr Leu Val Phe Gly Ala Leu Ser Asp Thr Phe Gly Arg
        275                 280                 285

Lys Thr Met Tyr Leu Leu Gly Ala Ala Ala Met Gly Val Val Ile Ala
    290                 295                 300

Pro Ala Phe Ala Leu Ile Asn Thr Gly Asn Pro Trp Leu Phe Met Ala
305                 310                 315                 320

Ala Gln Val Leu Val Phe Gly Ile Ala Met Ala Pro Ala Ala Gly Val
                325                 330                 335

Thr Gly Ser Leu Phe Thr Met Val Phe Asp Ala Asp Val Arg Tyr Ser
            340                 345                 350

Gly Val Ser Ile Gly Tyr Thr Ile Ser Gln Val Ala Gly Ser Ala Phe
        355                 360                 365

Ala Pro Thr Ile Ala Thr Ala Leu Tyr Ala Ser Thr Asn Thr Ser Asn
    370                 375                 380

Ser Ile Val Thr Tyr Leu Leu Ile Val Ser Ala Ile Ser Ile Val Ser
385                 390                 395                 400

Val Ile Leu Leu Pro Gly Gly Trp Gly Arg Lys Gly Ala Ala Ser Gln
                405                 410                 415

Leu Thr Arg Asp Gln Ala Thr Ser Thr Pro Lys Met Pro Asp Thr Glu
            420                 425                 430

Thr Phe Ser Thr Arg Thr Val Pro Asp Thr Ala Ala Ser Leu Arg Val
        435                 440                 445

Leu Asp Lys
    450
```

<210> 5
<211> 636
<212> PRT
<213> Rhodococcus corallina

<400> 5

```
Met Thr Asp Met Ser Asp His Asp Arg Thr Ser Tyr Asp Thr Asp Val
  1               5                  10                  15

Val Ile Val Gly Leu Gly Pro Ala Gly Gly Thr Ala Ala Leu Ala Leu
             20                  25                  30

Ala Ser Tyr Gly Ile Arg Val His Ala Val Ser Met Phe Pro Trp Val
         35                  40                  45
```

```
Ala Asn Ser Pro Arg Ala His Ile Thr Asn Gln Arg Ala Val Glu Val
     50                  55                  60

Leu Arg Asp Leu Gly Val Glu Asp Glu Ala Arg Asn Tyr Ala Thr Pro
 65                  70                  75                  80

Trp Asp Gln Met Gly Asp Thr Leu Phe Thr Thr Ser Leu Ala Gly Glu
                 85                  90                  95

Glu Ile Val Arg Met Gln Thr Trp Gly Thr Gly Asp Ile Arg Tyr Gly
            100                 105                 110

Asp Tyr Leu Ser Gly Ser Pro Cys Thr Met Leu Asp Ile Pro Gln Pro
        115                 120                 125

Leu Met Glu Pro Val Leu Ile Lys Asn Ala Ala Glu Arg Gly Ala Val
    130                 135                 140

Ile Ser Phe Asn Thr Glu Tyr Leu Asp His Ala Gln Asp Glu Asp Gly
145                 150                 155                 160

Val Thr Val Arg Phe Arg Asp Val Arg Ser Gly Thr Val Phe Thr Gln
                165                 170                 175

Arg Ala Arg Phe Leu Leu Gly Phe Asp Gly Ala Arg Ser Lys Ile Ala
            180                 185                 190

Glu Gln Ile Gly Leu Pro Phe Glu Gly Glu Leu Ala Arg Ala Gly Thr
        195                 200                 205

Ala Tyr Ile Leu Phe Asn Ala Asp Leu Ser Lys Tyr Val Ala His Arg
    210                 215                 220

Pro Ser Ile Leu His Trp Ile Val Asn Ser Lys Ala Gly Phe Gly Glu
225                 230                 235                 240

Ile Gly Met Gly Leu Leu Arg Ala Ile Arg Pro Trp Asp Gln Trp Ile
                245                 250                 255

Ala Gly Trp Gly Phe Asp Met Ala Asn Gly Glu Pro Asp Val Ser Asp
            260                 265                 270

Asp Val Val Leu Glu Gln Ile Arg Thr Leu Val Gly Asp Pro His Leu
        275                 280                 285

Asp Val Glu Ile Val Ser Arg Ser Phe Trp Tyr Val Asn Arg Gln Trp
    290                 295                 300

Ala Glu His Tyr Gln Ser Gly Arg Val Phe Cys Gly Gly Asp Ala Val
305                 310                 315                 320

His Arg His Pro Pro Ser Ser Gly Leu Gly Ser Asn Thr Ser Met Gln
                325                 330                 335

Asp Ala Phe Asn Leu Ala Trp Lys Ile Ala Phe Val Val Lys Gly Tyr
            340                 345                 350
```

```
Ala Gly Pro Gly Leu Leu Glu Ser Tyr Ser Pro Glu Arg Val Pro Val
        355                 360                 365

Gly Lys Gln Ile Val Ala Arg Ala Asn Gln Ser Arg Lys Asp Tyr Ala
    370                 375                 380

Gly Leu Arg Glu Trp Phe Asp His Glu Ser Asp Asp Pro Val Ala Ala
385                 390                 395                 400

Gly Leu Ala Lys Leu Lys Glu Pro Ser Ser Glu Gly Val Ala Leu Arg
                405                 410                 415

Glu Arg Leu Tyr Glu Ala Leu Glu Val Lys Asn Ala Glu Phe Asn Ala
            420                 425                 430

Gln Gly Val Glu Leu Asn Gln Arg Tyr Thr Ser Ser Ala Val Val Pro
        435                 440                 445

Asp Pro Glu Ala Gly Glu Glu Val Trp Val Arg Asp Arg Glu Leu Tyr
    450                 455                 460

Leu Gln Ala Thr Thr Arg Pro Gly Ala Lys Leu Pro His Ala Trp Leu
465                 470                 475                 480

Val Gly Ala Asp Gly Thr Arg Ile Ser Thr Leu Asp Val Thr Gly Lys
                485                 490                 495

Gly Met Met Thr Leu Leu Thr Gly Leu Gly Gly Gln Ala Trp Lys Arg
            500                 505                 510

Ala Ala Ala Lys Leu Asp Leu Pro Phe Leu Arg Thr Val Val Val Gly
        515                 520                 525

Glu Pro Gly Thr Ile Asp Pro Tyr Gly Tyr Trp Arg Arg Val Arg Asp
    530                 535                 540

Ile Asp Glu Ala Gly Ala Leu Leu Val Arg Pro Asp Gly Tyr Val Ala
545                 550                 555                 560

Trp Arg His Ser Ala Pro Val Trp Asp Asp Thr Glu Ala Leu Thr Ser
                565                 570                 575

Leu Glu Asn Ala Leu Thr Ala Val Leu Asp His Ser Ala Ser Asp Asn
            580                 585                 590

Gly Asn Pro Ser Gly Thr Asn Glu Pro Gln Tyr Ser Thr Arg Ala Val
        595                 600                 605

Pro Ile Val Val Pro His Val Thr Ala Glu Asp Ala Ala Pro Ala Ser
    610                 615                 620

Ala Thr Arg Thr Thr Thr Val Glu Gly Glu Asn Arg
625                 630                 635
```

<210> 6
<211> 289
<212> PRT
<213> Rhodococcus corallina

<400> 6

```
Met Thr Arg Pro Tyr Thr Ser Val Trp Asp Asp Leu Asn Gln Val Glu
  1               5                  10                  15

Phe Ser Gln Gly Phe Ile Gln Ala Gly Pro Tyr Arg Thr Arg Tyr Leu
             20                  25                  30

His Ala Gly Asp Ser Ser Lys Pro Thr Leu Ile Leu Leu His Gly Ile
         35                  40                  45

Thr Gly His Ala Glu Ala Tyr Val Arg Asn Leu Arg Ser His Ser Glu
     50                  55                  60

His Phe Asn Val Trp Ala Ile Asp Phe Ile Gly His Gly Tyr Ser Thr
 65                  70                  75                  80

Lys Pro Asp His Pro Leu Glu Ile Lys His Tyr Ile Asp His Val Leu
                 85                  90                  95

Gln Leu Leu Asp Ala Ile Gly Val Glu Lys Ala Ser Phe Ser Gly Glu
            100                 105                 110

Ser Leu Gly Gly Trp Val Thr Ala Gln Phe Ala His Asp His Pro Glu
        115                 120                 125

Lys Val Asp Arg Ile Val Leu Asn Thr Met Gly Gly Thr Met Ala Asn
    130                 135                 140

Pro Gln Val Met Glu Arg Leu Tyr Thr Leu Ser Met Glu Ala Ala Lys
145                 150                 155                 160

Asp Pro Ser Trp Glu Arg Val Lys Ala Arg Leu Glu Trp Leu Met Ala
                165                 170                 175

Asp Pro Thr Met Val Thr Asp Asp Leu Ile Arg Thr Arg Gln Ala Ile
            180                 185                 190

Phe Gln Gln Pro Asp Trp Leu Lys Ala Cys Glu Met Asn Met Ala Leu
        195                 200                 205

Gln Asp Leu Glu Thr Arg Lys Arg Asn Met Ile Thr Asp Ala Thr Leu
    210                 215                 220

Asn Gly Ile Thr Val Pro Ala Met Val Leu Trp Thr Thr Lys Asp Pro
225                 230                 235                 240

Ser Gly Pro Val Asp Glu Ala Lys Arg Ile Ala Ser His Ile Pro Gly
                245                 250                 255

Ala Lys Leu Ala Ile Met Glu Asn Cys Gly His Trp Pro Gln Tyr Glu
            260                 265                 270
```

```
Asp Pro Glu Thr Phe Asn Lys Leu His Leu Asp Phe Leu Leu Gly Arg
        275                 280                 285

Ser
```

<210> 7
<211> 314
<212> PRT
<213> Rhodococcus corallina

<400> 7

```
Met Pro Val Ala Leu Cys Ala Met Ser His Ser Pro Leu Met Gly Arg
  1               5                  10                  15

Asn Asp Pro Glu Gln Glu Val Ile Asp Ala Val Asp Ala Ala Phe Asp
            20                  25                  30

His Ala Arg Arg Phe Val Ala Asp Phe Ala Pro Asp Leu Ile Val Ile
        35                  40                  45

Phe Ala Pro Asp His Tyr Asn Gly Val Phe Tyr Asp Leu Leu Pro Pro
    50                  55                  60

Phe Cys Ile Gly Ala Ala Ala Gln Ser Val Gly Asp Tyr Gly Thr Glu
65                  70                  75                  80

Ala Gly Pro Leu Asp Val Asp Arg Asp Ala Ala Tyr Ala Val Ala Arg
                85                  90                  95

Asp Val Leu Asp Ser Gly Ile Asp Val Ala Phe Ser Glu Arg Met His
            100                 105                 110

Val Asp His Gly Phe Ala Gln Ala Leu Gln Leu Leu Val Gly Ser Ile
        115                 120                 125

Thr Ala Val Pro Thr Val Pro Ile Phe Ile Asn Ser Val Ala Glu Pro
    130                 135                 140

Leu Gly Pro Val Ser Arg Val Arg Leu Leu Gly Glu Ala Val Gly Arg
145                 150                 155                 160

Ala Ala Ala Lys Leu Asp Lys Arg Val Leu Phe Val Gly Ser Gly Gly
                165                 170                 175

Leu Ser His Asp Pro Pro Val Pro Gln Phe Ala Thr Ala Pro Glu Glu
            180                 185                 190

Val Arg Glu Arg Leu Ile Asp Gly Arg Asn Pro Ser Ala Ala Glu Arg
        195                 200                 205

Asp Ala Arg Glu Gln Arg Val Ile Thr Ala Gly Arg Asp Phe Ala Ala
    210                 215                 220

Gly Thr Ala Ala Ile Gln Pro Leu Asn Pro Glu Trp Asp Arg His Leu
225                 230                 235                 240
```

```
Leu Asp Val Leu Ala Ser Gly Asp Leu Glu Gln Ile Asp Ala Trp Thr
                245                 250                 255

Asn Asp Trp Phe Val Glu Gln Ala Gly His Ser Ser His Glu Val Arg
            260                 265                 270

Thr Trp Ile Ala Ala Tyr Ala Ala Met Ser Ala Ala Gly Lys Tyr Arg
        275                 280                 285

Val Thr Ser Thr Phe Tyr Arg Glu Ile His Glu Trp Ile Ala Gly Phe
    290                 295                 300

Gly Ile Thr Thr Ala Val Ala Val Asp Glu
305                 310
```

<210> 8
<211> 289
<212> PRT
<213> Rhodococcus corallina

<400> 8

```
Met Thr Ser Val Arg Pro Cys Ser Pro Ser Val Asn Ala Gly Trp Ser
  1               5                  10                  15

Val Gly Arg Lys Thr Ser Ser Pro Thr Ser Pro Ser Thr Ser Gln Leu
             20                  25                  30

Val Ser Arg Asn Ala His Gly Pro Thr Ser Arg Ala Gly His Arg Gly
         35                  40                  45

Gln Pro Arg His Arg Gly Gly His Arg Arg Cys Gly Gly Arg Leu Arg
     50                  55                  60

Cys Arg Arg Asn Arg Pro Leu Arg Ile Arg Ser Asp Gly Arg Arg Cys
 65                  70                  75                  80

Gly Val Asp Gly Ile Thr Ala Ala Gly Gly Leu Ala Ala Ala Val Gln
                 85                  90                  95

Ala Asp Leu Ser Arg Pro Glu Gly Pro Glu Glu Leu Met Arg Glu Phe
            100                 105                 110

Asp Ser Ala Leu Asp Gly Leu Gly Leu Asp Arg Gly Leu Asp Ile Leu
        115                 120                 125

Val Asn Asn Ala Gly Ile Ser Arg Arg Gly Ala Leu Glu Arg Val Thr
    130                 135                 140

Val Glu Asp Phe Asp Arg Leu Val Ala Leu Asn Gln Arg Ala Pro Phe
145                 150                 155                 160

Phe Val Thr Arg His Ala Leu Pro Arg Met His Asp Gly Gly Arg Ile
                165                 170                 175

Val Asn Ile Ser Ser Gly Ser Ala Arg Tyr Ala Arg Pro Asp Val Ile
            180                 185                 190
```

Ser Tyr Ala Met Thr Lys Gly Ala Ile Glu Val Leu Thr Arg Ala Leu
195 200 205

Ala Val Asp Val Gly Glu Arg Gly Ile Thr Ala Asn Ala Val Ala Pro
210 215 220

Ala Ala Leu Asp Thr Asp Met Asn Ala His Trp Leu Arg Gly Asp Asp
225 230 235 240

His Ala Arg Thr Thr Ala Ala Ser Thr Thr Ala Leu Arg Lys Leu Ala
245 250 255

Thr Ala Glu Asp Ile Ala Ala Ile Val Ala Phe Leu Val Ser Ala Ala
260 265 270

Ala Gly Ala Ile Thr Gly Gln Val Ile Asp Ala Thr Asn Gly Asn Arg
275 280 285

Leu

<210> 9
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 9
cgctgatttg tattgtctg 19

<210> 10
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 10
gacttccatt gttcattcc 19

<210> 11
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 11
aaaagacgtc ggtgcgaata agggacagtg 30

<210> 12
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 12
aaaagacgtc acaaaacagc agggaagcag 30